

SOFT-PART PRESERVATION OF BEETLES IN TERTIARY AMBER FROM THE DOMINICAN REPUBLIC

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ABSTRACT. Exceptional soft-part preservation of the internal anatomy of beetles (Nitidulidae and Cantharidae) is described from Tertiary amber, for the first time from the Dominican Republic. A variety of tissues is preserved, including elements of the nervous, digestive, respiratory, locomotory and sensory systems.

AMBER is celebrated for the exquisite preservation of its fauna, especially the exoskeletons of fossil insects. However, the common assumption that these fossils are nothing more than empty moulds (Brues 1951) lined only with the decomposed vestiges of soft tissue has been little questioned. Described here is the preservation of soft-parts in beetles in Tertiary amber from the Dominican Republic. This material expands our knowledge of soft-part preservation in two ways. First, soft-part preservation is shown to occur apparently in the absence of any special circumstances; previously Poinar and Hess (1982, 1985) described preservation in an amber inclusion in which resin had unusually penetrated the body cavity. Second, extensive areas of tissue are described including a section through the body, in contrast to the 12 μ m wide strip documented by Poinar and Hess (1982, 1985) and flight muscle described by Henwood (1992). Additionally, as a practical consideration, the soft-part preservation described is from amber inclusions which, compared to Baltic amber inclusions, are considerably more numerous both in the field and in collections; hence the material is more easily available to palaeontologists.

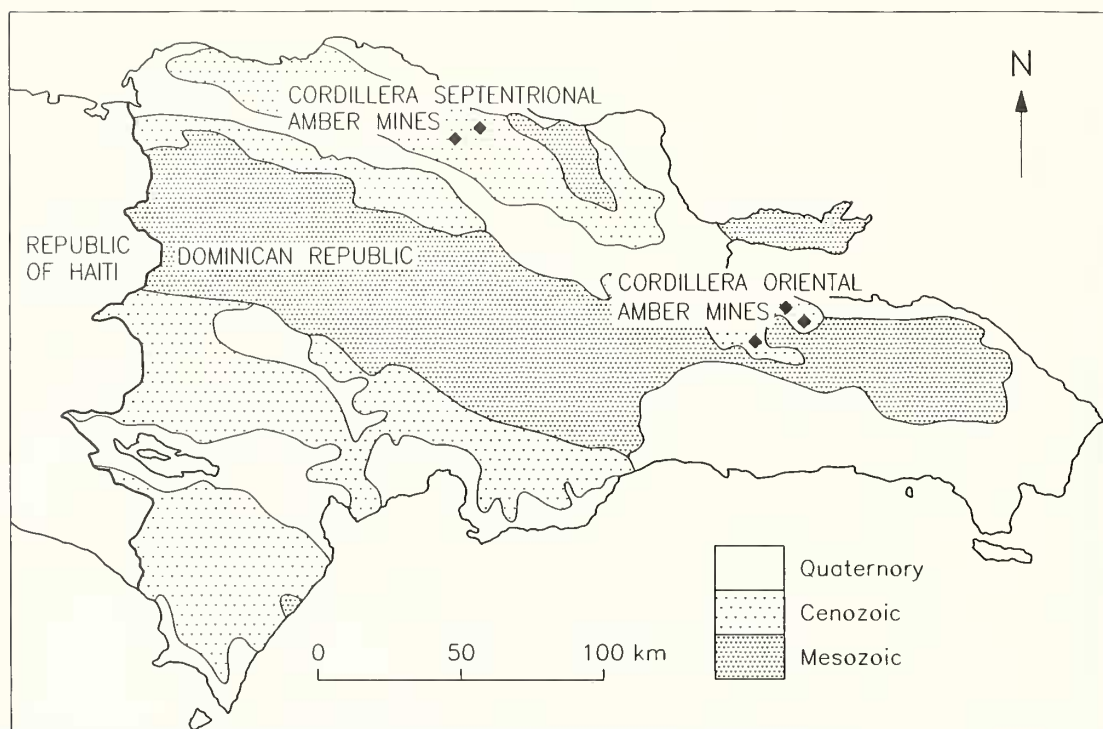
Amber fossils include a range of terrestrial organisms among which are relatively soft-bodied organisms otherwise scarce in the fossil record. For example, amber fossils account for a substantial part of the fossil record of insects, the most numerous animals presently on Earth (for example see Crowson 1981; Keilbach 1982). The variety of inclusions in Baltic Eocene amber spans almost every insect order, and also includes pseudoscorpions and a reptile (Larsson 1978). Amber inclusions are increasingly useful to palaeontologists because they are known from a wide range of geological ages. For example, Lower Cretaceous amber is known from Lebanon (Schlee and Dietrich 1970) and Siberia (Alekseyev and Rasnitsyn 1981) with insect inclusions representing a fauna contemporaneous with the origin of angiosperms (Friis *et al.* 1987). Organisms fossilized in amber thus make an important contribution to the fossil record.

The preservation of soft tissue in insect amber fossils compares favourably with that of several Konservat-Lagerstätten which yield histological information. In contrast to the preservation of the Messel brown coals, where soft tissue preservation was observed 'only in a few small areas of the smaller animals' (Voigt 1988), it appears that insects fossilized in amber can be preserved in their entirety. Furthermore whereas the preservation of histological detail in 'conventional fossils' usually results from mineral replacement (for example the phosphate-replaced muscles of fish from the Cretaceous Santana Formation of Brazil (Martill 1990) in which muscle nuclei can be seen), the actual tissue appears to be preserved in amber fossils.

PREVIOUS WORK

The common assumption that amber fossils are empty moulds was brought into question by Poinar and Hess (1982, 1985) who examined a mycetophilid fly from Baltic amber. Penetration of the body

cavity by resin, which is usually excluded by the impermeable cuticle, contributed to the unusual preservational circumstances in this case. Poinar and Hess (1982, 1985) showed that only a 12 μm wide strip of tissue, adjacent to the cuticle, was preserved. Within a few areas of this tissue they reported preserved cell ultrastructure including nuclei and apparent lipid droplets, ribosomes, and elongate bodies resembling mitochondria. As this tissue came from the abdomen, it was suggested that this tissue represented epidermis and associated muscle.



TEXT-FIG. 1. Geological sketch map of the Dominican Republic showing the location of mines in the two principal amber-producing areas, the central Cordillera Septentrional and the eastern Cordillera Oriental (after Saunders *et al.* 1986).

Henwood (1992) documented ultrastructural preservation in an amber-fossilized dipteran fly (Suborder Brachycera; Epididae) from the Dominican Republic in which there were no exceptional preservational circumstances. Transmission electron microscopy of the flight muscle showed transverse sections through bundles of longitudinal myofibrils, between which mitochondria are densely packed (Henwood 1992). The mitochondria were sufficiently well preserved that even their cristae could be distinguished, and were frequently preserved in parallel stacks. Tracheoles of less than 1 μm diameter were present, intimately associated with the muscle fibres.

AMBER FROM THE DOMINICAN REPUBLIC

Scientific attention was first directed to the Dominican Republic's extensive amber deposits and their numerous biotic inclusions by Sanderson and Farr (1960). Since then, many workers have described the flora and fauna of Dominican amber. In addition to abundant insects and arachnids, mammalian hair (Poinar 1988), a leptodactylid frog (Poinar and Cannatella 1987) and a gilled mushroom (Poinar and Singer 1990) have been recorded.

Several general points about the depositional environment of amber emerge from a literature survey and the fieldwork I have undertaken. Not unexpectedly amber is almost always associated with lignite. All of the amber-bearing sediments studied represent some kind of flood, (for example overbank flood) or slump deposit, in which the removal of water which had acted as the transporting medium caused the deposition of the sediment load, since not all amber, with a specific gravity ranging from 1 to 1.25 (Langenheim 1964), will settle out in water.

There are two principal amber-bearing localities in the Dominican Republic; the central Cordillera Septentrional and the Cordillera Oriental (Text-fig. 1).

Geology of the Cordillera Septentrional amber deposits

The Cordillera Septentrional is a west-northwest trending mountain belt extending along the Atlantic coast of the Dominican Republic (Text-fig. 1). It is composed of three major stratigraphical groups: a Late Cretaceous to Early Eocene basement of volcanic rock; a thick succession of Eocene to Middle Miocene sedimentary rocks which have been referred to by a variety of names (the El Mamey belt, the Altamira Formation, the La Toca Formation and the Las Larvas Formation; Zoeten 1988); and an Upper Miocene cap of carbonates, mostly marls. The amber is found in a limited number of horizons within the middle formation; most frequently it occurs in a laminated blue-grey biotite siltstone with a rich organic content, and infrequently in a conglomerate or a fine sandstone. The pieces of amber are found either associated with, or sometimes within, lignite. Amber clasts are found in a variety of forms, frequently with tapered edges and processes consistent with their origin as resinous tree exudate. The distinct and relatively sharp edges of the amber clasts found in the Cordillera Septentrional may indicate that reworking is insignificant.

Alternative hypotheses, based on sedimentary evidence, have been proposed for the depositional environment. Eberle *et al.* (1982) suggested that the presence of lignite seams, locally associated with amber, as well as the thick and extensive orthoconglomerates, indicated a fan-related shallow water environment. If this alluvial fan model is valid, the conglomerates would represent the channel section of the fan, lignite- and amber-rich beds would have been deposited in the interchannel flats, and marly portions of the shale and sandstone formation would represent the distal fan base environment. Less convincingly, Redmond (1982) interpreted a massive sandstone in the sequence to indicate a mid-fan environment beyond the channels, where sand from the channels prograded over the amber-bearing units. In the main body of the sediments, fossils are rare and do not provide satisfactory biostratigraphical information.

Geology of the Cordillera Oriental amber deposits

The amber deposits of the eastern Cordillera Oriental region of the Dominican Republic occur in a series of small valleys around the town of El Valle. These small valleys are eroded into volcanic rocks which constitute the basement on which the amber-bearing sediments lie (Brouwer and Brouwer 1982). Regional tectonic studies have suggested uplift, possibly due to intrusion and block faulting (with predominantly vertical displacements), consistent with the larger scale tectonic setting of the region (Lewis and Draper 1990). The amber-bearing sequences are composed principally of poorly-sorted claystones, calcarenites, and calcareous and carbonaceous mudstones. In most cases the sediments are horizontal, but in the La Cumbre mines south of El Valle they are inclined.

The largest amount of amber is yielded by a single bed of carbonaceous calcareous mudstone which also contains discontinuous lenses of lignite. The rounded nature of amber clasts found in the Cordillera Oriental, in contrast to the apparently unworn nature of clasts from the Cordillera Septentrional, perhaps indicates that amber from the former locality is more extensively reworked than that from the latter. In the vicinity of El Valle this sequence has been interpreted as a cyclothem deposited within a shallow depositional basin such as occurs on swampy coasts with repeated fluctuations of sea level (Brouwer and Brouwer 1982).

AGE OF THE AMBER

As amber cannot be dated, the age of the sediments gives only a minimum for the amber. However, since the Cordillera Septentrional amber is little reworked, the amber-bearing sediments probably reflect the age of the amber quite closely. Additionally, the absence of marine, biostratigraphically useful, fossils in both the Cordillera Septentrional and Cordillera Oriental sediments complicates the situation. In the central Cordillera Septentrional, microfaunal analysis of the amber-bearing

sediments from Palo Alto mine, indicates a mid Oligocene to earliest Miocene age (23–30 Ma) (Baroni-Urbani and Saunders 1980). Eberle *et al.* (1980) regarded the amber-bearing sediments as part of the Altamira facies of the El Mamey Formation which they dated as Later Eocene (35–40 Ma). Cepek (as reported by Schlee 1990), dated the amber deposits as Middle Eocene (50 Ma).

Less credible is the Upper Eocene (35–40 Ma) age estimated for sediments from La Toca mine in the central Cordillera Septentrional (Lambert *et al.* 1985) based upon relative differences in nuclear magnetic resonance spectra calibrated with the earliest Miocene age of the Palo Alto sediments which was biostratigraphically determined by Baroni-Urbani and Saunders (1980). However, the vigorous faulting of the La Toca sediments provides an alternative, and more parsimonious, explanation of the spectral differences. Thin sections of the sediments from Palo Alto and La Toca are very similar notwithstanding the deformation in the La Toca section.

Amber from the Cordillera Oriental is found in two formations which are probably of the same age. Amber at El Valle is found in the Yanigua Formation of Middle to Early Miocene age (14–23 Ma). In the vicinity of East Cibao, amber is mined from the Los Guayuyos Formation (the upper part of the Cercado Formation) of late Early Miocene age (16–23 Ma) (Brouwer and Brouwer 1982).

Future determination of more precise age ranges for the amber fossils from the Dominican Republic will depend on two developments. First, the expansion of chemical ‘fingerprinting’ techniques, such as pyrolysis gas chromatography, by which the provenance of a particular piece of amber can be pinpointed. Second, an improved knowledge of the geology and therefore ages of the amber-bearing strata of the Dominican Republic.

MATERIALS AND METHODS

Provenance of the material

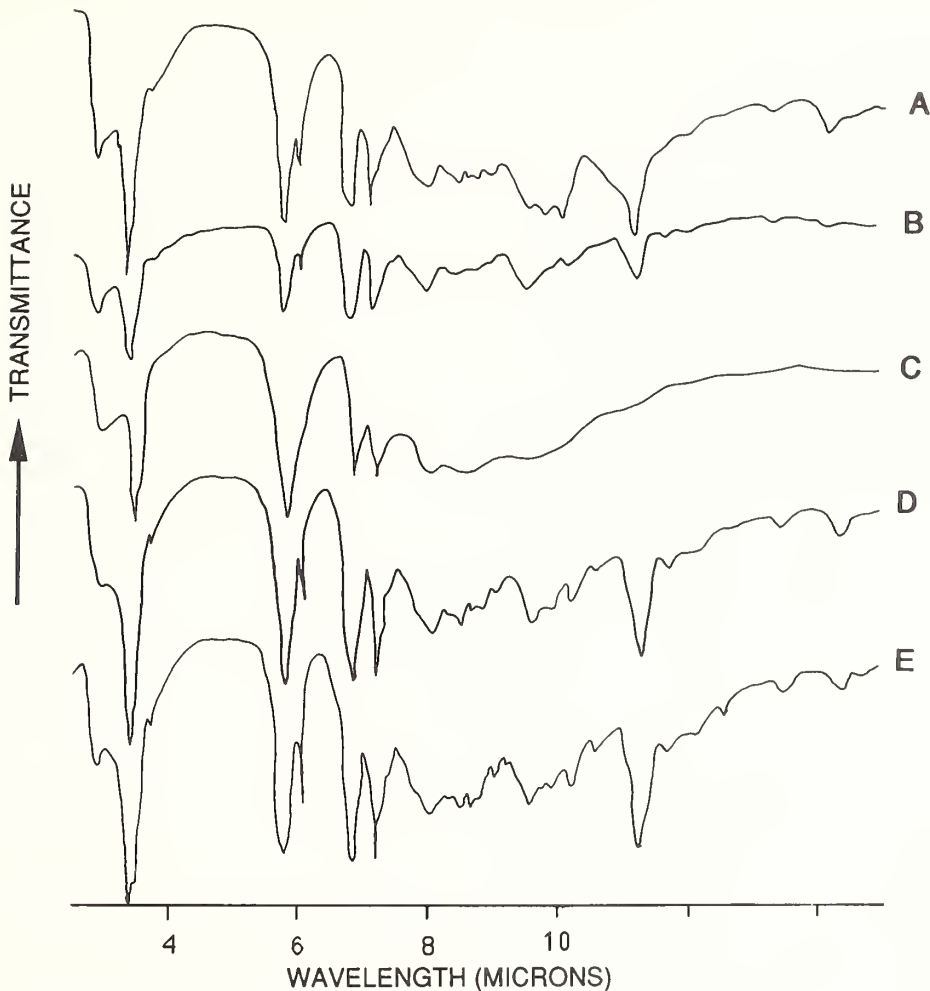
The amber fossils in this study were purchased from J. Brodzinsky of Santo Domingo in the Dominican Republic. Unfortunately, no reliable locality information was available with the specimens because inclusions are not visible until the amber is polished; amber from different sources is mixed for working and polishing. Infrared spectra cannot provide any independent information on the age of these individual pieces of amber, but similarity between spectra can indicate provenance. An infrared spectral comparison between amber of known provenance and the purchased samples (Text-fig. 2) suggests that the latter are from Palo Alto in the Cordillera Septentrional. They share a unique pattern of: (1) a marked drop in transmittance at 11–11.2 μm wavelength, (2) two successively greater peaks between 9.5 and 10.2 μm , and (3) another separate peak around 10.5 μm . This indicates that these ambers are probably Middle Oligocene to lower Early Miocene in age (23–30 Ma). The authenticity of the amber samples examined here was also confirmed by the close similarity of their infrared spectra to published spectra (Langenheim and Beck 1968), and to spectra from genuine amber I have collected from these localities.

Methods

Two methods were used to examine the preservation of the internal material: either sections were cut using an annular saw, or the body of the insect was fractured to expose its contents. One specimen (a cantharid) was serially sectioned, at c. 250 μm intervals (Sedgwick Museum, Cambridge, specimens SM X.23256–X.23257). In the second method, a slow-speed diamond saw was used to cut away the amber surrounding the insect (a nitidulid) and the body of the insect was then fractured (SM X.23254, X.23258). The fractured sections were gold coated and examined in a Jeol JSM820 scanning electron microscope at a range of operating voltages.

PRESERVATION

The external preservation of the beetles is good, with no apparent damage to the exoskeleton. The wings and elytra of both specimens were folded in a resting position. Both beetles are small; the cantharid c. 5 mm long, and the nitidulid c. 2.5 mm long. The enclosing amber is pale yellow, and that containing the cantharid beetle also contained numerous tiny bubbles (mostly less than 0.5 mm diameter). Some of these bubbles themselves contain bubbles, indicating the presence of at least two substances, probably air and water, both of which have previously been documented as amber inclusions (Berner and Landis 1988; Schlee 1980).



TEXT-FIG. 2. Infrared spectra of amber samples from the Dominican Republic. A, amber collected by the author at El Valle mine, Cordillera Oriental; B, published reference spectra (Langenheim and Beck 1968), Palo Alto, Cordillera Septentrional; C, published reference spectra (Langenheim and Beck 1968), Palo Quemado, Cordillera Septentrional; D, SM X.23255-6, purchased amber containing a cantharid beetle, from the Dominican Republic, provenance unknown; E, SM X.23254, purchased amber containing a nitidulid beetle, Dominican Republic, provenance unknown.

Internal anatomy is preserved with fidelity. Sections through the whole insect show that the body remains essentially as in life, full of soft tissues (Pl. 1, fig. 1; Text-fig. 3). These tissues include material from a variety of organ systems: locomotory (Pl. 1, fig. 2), digestive (Pl. 1, fig. 3; Text-fig. 3A), respiratory (Pl. 1, fig. 4; Text-fig. 3B), nervous (Pl. 1, figs 5 and 6) and sensory (Pl. 1, figs 7 and 8). Tissues are preserved in three dimensions, together with their relative positions and interconnections as within the living body. Furthermore it is the actual tissue which is believed to be preserved, enabling preservation to extend to the ultrastructure of cell organelles (Henwood 1992). The sections do not appear to have decayed since cutting in February 1991.

Muscle fibres are widely preserved, and flight muscle is exceptionally well preserved (Pl. 1, fig. 2). A complete dorso-ventral transverse section through the thorax is seen in a cantharid beetle (Pl. 1, fig. 1). The flight muscle is a dense regular structure with closely packed mitochondria defining myofilament channels, although the myofilaments themselves appear to have disintegrated. The broken surfaces of the mitochondria have

indentations which may represent the routes of T-tubules in the original muscle (Smith 1966). All extant coleopterans have asynchronous muscle (Pringle 1980) capable of contracting faster than the rate of receipt of nerve impulses. Unlike other insect orders, for example Diptera, extant Coleoptera have the T-tubules positioned irregularly relative to the bands of the sarcomeres (Smith 1966); their spacing cannot therefore indicate sarcomere dimensions.

The myofibrils of amber-preserved coleopteran flight muscle are about half the diameter of those from fresh insect material, e.g. the coleopteran *Tenebrio molitor* (see Smith 1961). Comparison of dipteran flight muscle myofibril and mitochondria ultrastructure using transmission electron microscopy, together with decay experiments, indicated that preservation in amber was accompanied by approximately 50 per cent shrinkage (Henwood 1992). This suggests that the change in the size of myofibril observed here is also the result of shrinkage during preservation rather than evolutionary change in coleopteran myofibril size.

Associated with the flight muscle are large numbers of tracheae (Pl. 1, fig. 4). Encircling these, either as spirals or rings, are thickened bands (taenidia). Large tracheae (c. 60 μm diameter) repeatedly divide into smaller tracheoles (less than 1 μm diameter) to make intimate contact with the respiring tissues. Minute tracheoles (less than 0.3 μm diameter), are preserved *in situ* between individual fibrils of the flight muscle, and are seen to run across a flight muscle fibre (Pl. 1, fig. 2).

The proventriculus, the posterior region of the foregut, which functions as an accessory chewing apparatus and as a valve which controls the movement of food, is preserved (Text-fig. 3A) as is the foregut (Text-fig. 3B). The former is divided into a number of lobes, eight in this example, and through the middle of each runs a thin, finely sculptured ridge. Groups of several tens of similarly sized muscle fibres are attached to 'teeth' which project into the lumen of the proventriculus (Pl. 1, fig. 3). These bundles of muscle fibres, of 2–3 μm diameter, attach the proventricular teeth to the inner surface of the proventriculus. The proventricular teeth are spatulate, c. 30 μm wide by 40 μm high, with a narrow crenation along their upper edge. The body of the tooth is a composite of many collagenous fibres, each less than 0.5 μm wide. These teeth probably functioned in the mastication of food (Balfour-Browne 1944). The structure of the proventricular teeth suggests that early Tertiary nitidulids, like their extant relatives, had a relatively soft diet, such as fungi or decaying plant matter (R. Northfield, pers. comm.).

Elements of the nervous system can be tentatively identified, in conjunction with sensory organs, including glial cells and the axons of neurons. Frequently it is the outer fibrous sheath of glial cells, c. 2 μm diameter, that is visible. Shrinkage has been documented in flight muscle from an insect fossilized in amber (Henwood 1992) suggesting that glial cells and axons may also have been reduced in size during preservation. Internal views of the sensillae bases show a transverse section of what is believed to be the outer dendrite segment that runs to the tip of the sensillar hair. The sensory cell, and the thecogen, trichogen and tormogen cells which envelop it in the sensilla of extant insects (Keil and Steinbrecht 1982) are not visible. Glial cells with ensheathed axons are believed to run from the base of each sensillum (Pl. 1, fig. 5). Below the many ommatidia of the compound eyes, axons are visible across what is believed to be the basement of the retinula (Pl. 1, fig. 8). A section through the head (Pl. 1, fig. 7) shows both eyes and some brain tissue. Unfortunately much of this section, including most of the brain, is missing and is believed to have been lost as a result of the use of the annular saw (see below).

EXPLANATION OF PLATE 1

Soft tissues preserved in beetles from Oligocene amber from the Dominican Republic.

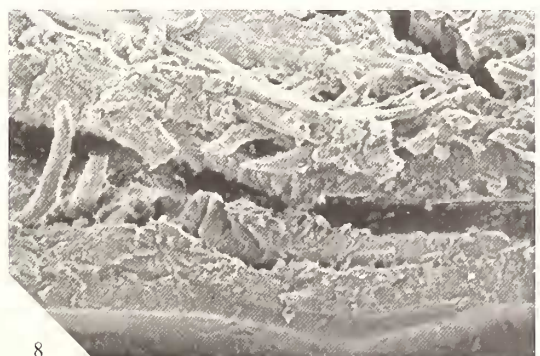
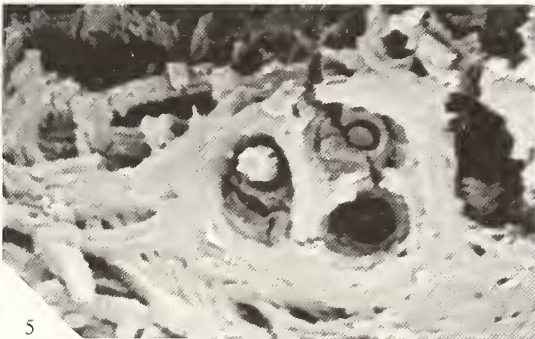
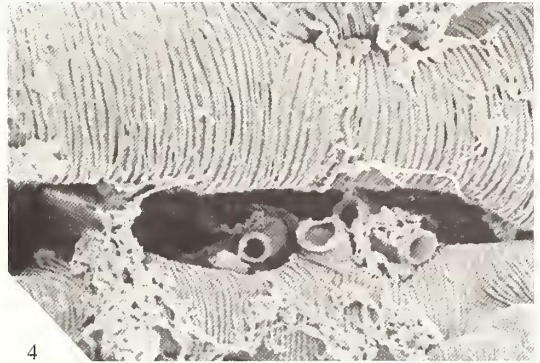
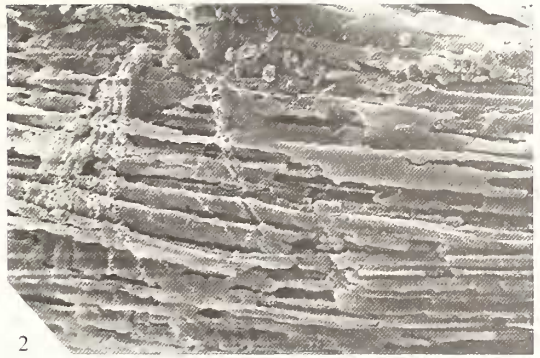
Figs 1–2, 4, 6. Cantharid beetle, SM X.23256. 1, view of a dorso-ventral transverse section through the thorax, $\times 60$. 2, flight muscle with densely packed parallel fibrils and tracheoles running across the surface, $\times 1320$. 4, axial trachea and smaller tracheoles showing characteristic annular thickenings (taenidia) servicing the flight muscle, $\times 9000$. 6, external view of a sensillar field with the hair-like projections missing from all but one of the sensilla, $\times 5100$.

Fig. 3. Nitidulid beetle, SM X.23254; proventricular teeth with attachment musculature from the inner surface of the proventriculus, $\times 670$.

Fig. 5. Cantharid beetle, SM X.23257; internal surface of a ventral midline sensilla field showing basal glial cells and the ensheathed axons, $\times 2700$.

Figs 7–8. Cantharid beetle, SM X.23258. 7, transverse section through the head with the compound eyes visible although much of the brain tissue is missing, $\times 95$. 8, retinula from the base of the ommatidia showing neurons, $\times 2700$.

All figures are scanning electron micrographs.





TEXT-FIG. 3. Soft-part preservation shown in part and counterpart of fracture surface across a nitidulid beetle preserved in Oligocene amber from the Dominican Republic. A, the proventriculus (posterior region of the foregut) showing detail of sculpture on lobes; SM X.23254, $\times 260$. B, counterpart showing foregut with encircling tracheal supply; SM X.23258, $\times 170$. Both figures are scanning electron micrographs.

Part of the outer surfaces of both compound eyes is visible in a transverse section of a cantharid beetle. The eyes are composed of approximately 60 identically-sized ommatidia. There is little convexity of the individual ommatidia which suggest a diurnal habit (Crowson 1981). The outer corneal surfaces of the ommatidia are fairly smooth; no corneal tubercles or setae between the ommatidia are developed. A section through the ommatidia is not exposed in any of the material described here; no comment can therefore be made on their ultrastructure or state of preservation.

COMPOSITION OF THE MID-TERTIARY ATMOSPHERE

A comparison of amber-preserved dipteran flight muscle and modern dipteran flight muscle decaying under a variety of conditions indicates that 50 per cent is a conservative estimate of the myofibril shrinkage accompanying amber preservation (Henwood 1992). A reconstruction of the life dimensions of the flight muscle from Eocene specimens, estimated to have shrunk by about 50 per cent through preservation, suggests that the Eocene atmospheric oxygen level was higher than the present day. This is because the oxygen supply to the flight muscle from the axial trachea

depends on diffusion (Weis-Fogh 1964), and the minimum atmospheric oxygen composition for flight muscle function is related to the diameter of the flight muscle (Alexander 1979). This indication of higher atmospheric oxygen levels in the Eocene than today independently corroborates isotope and climate-modelling evidence of declining atmospheric oxygen through the Cenozoic (Shackleton 1985; Klump and Garrels 1986; Berner and Canfield 1989).

Alternative evolutionary explanations of the observed difference in flight muscle are discounted for two reasons. First, to explain the changes physiologically it is necessary to assume that Tertiary Coleoptera were more metabolically efficient than their Recent counterparts; this seems unlikely (there is no evidence to support this). Second, the tracheole diameters of Tertiary and Recent specimens (Richards 1951) are identical, and as they are lined by cuticle, are unlikely to shrink during preservation. Since oxygen supply to the flight muscle is limited by diffusion (Chapman 1982), to explain flight muscle changes by increased oxygen diffusion means proposing a different, more efficient, Tertiary tracheal supply unknown in Recent Coleoptera; again, there is no evidence to support this supposition.

DISCUSSION

At this level of resolution, little anatomical difference between extant and fossilized beetles can be identified. For several reasons this is as expected. Primary amongst these is the fact that amber-fossilized insects from the Dominican Republic are, at most, Middle Eocene in age (Schlee 1990) whereas the majority of extant coleopteran superfamilies are believed to be in the fossil record by the Middle Cretaceous (Crowson 1981). Furthermore the majority of Tertiary amber beetles, at least in the well-studied Baltic material, are very largely attributable to extant genera (Crowson 1981).

Technical considerations additionally constrain precise comparisons between extant and amber-fossilized material; amber-fossilized insects cannot yet be reconstructed with sufficient refinement. Problems have arisen in the preparation of insects for SEM examination. Several of the sections cut on the annular saw were empty, except for insubstantial layers of tissue around the edge. In this instance, this is believed to be the result of the fragile internal tissue being destroyed during cutting. Three observations support this explanation. First, when serial sections are unloaded from the saw onto SEM stubs, some material, similar in texture to that seen within the insects, can be seen displaced between sections. Second, in some empty sections, tracheae occur that are sharply broken off indicating that they did extend farther, and their distal ends were lost through breakage rather than decay which would not result in a sharp edge. Third, hollow sections are encountered adjacent to sections full of soft-parts; although preservation doubtless varies between individuals and maybe within individuals, it seems most unlikely that, within millimetres, the variation is so extreme. Loss of material is not believed to occur in fractured specimens, but this technique has the disadvantages of being both less precise and leaving more internal anatomy obscured.

The availability of the information necessary for detailed comparison between fossil and extant individuals is dependent on improved preparation of amber fossil for SEM examination. Controlled fracturing of fossil material is, to date, the most promising approach; unexposed soft-parts may be revealed by progressive, repeated fracturing of the fossil after thorough examination and photographic records have been made.

Preservational mechanism

Decay by microbes, fungi and bacteria, is the principal cause of the loss of information in the fossil record (Allison 1988); preservational mechanisms act to limit decay. Three suggested explanations for the preservation of insects in amber have appeared in the literature: (1) anaerobic conditions, (2) antibiotic action of resin, and (3) dehydration (Butterfield 1990). Of these only the first can be discounted as an important factor because amber is not completely airtight (Hopfenberg *et al.* 1988; Horibe and Craig 1988). The majority of evidence now is against the claim of Berner and Landis (1988) that amber was airtight. Furthermore anaerobic conditions do not necessarily impede biological decay (Allison 1988; Kidwell and Baumiller 1990; Penderson and Calvert 1990).

The evidence available suggests that both the antibiotic action of tree resin and dehydration may act to slow or halt the progress of decay. Documentation of the antibiotic properties of tree resin is rare although many anecdotal examples are cited (Poinar and Hess 1985). As yet there are scant data on the antibiotic properties of the resin of *Hymenaea courbaril*, the tree suggested as the botanical source of amber from the Dominican Republic (Hueber and Langenheim 1986). However, the oxide of one component of *H. courbaril* resin is known to have fungicidal properties (Arrhenius and Langenheim 1983).

Dehydration prevents decay because water, the medium in which all organic reactions occur, is removed. The importance of dehydration in the soft-part preservation of insects fossilized in amber has been indicated by comparison between decay sequences under various experimental conditions and amber-fossilized tissue (Henwood 1992). Water may be removed from insects embedded in tree resin by osmosis. However, such a dehydration model for the preservation of insects may be an oversimplification. The lack of an aqueous phase has also been suggested as a preservational cause for organisms preserved in tar seeps and mummification (Butterfield 1990), although preservation by mummification may be more complex. In preservational style, and possibly process too (see below), the preservation of insects in amber resembles the preservation of Egyptian mummies although these are only several thousand years old.

Comparison of the processes by which the preservation of Egyptian mummies was achieved with that of insects fossilized in amber is informative, especially in the relative roles of dehydration and antibiotic properties of resin. The highest fidelity of preservation in Egyptian mummies occurs in conjunction with widespread use of resin, both filling the body cavities and impregnating the outer bandages (Cockburn *et al.* 1975). In the more expensive mummifications, which presumably were known to result in improved preservation, the bandaging was preceded by an approximately seventy-day dehydration period of the body. Water was chemically extracted by packing the body cavity with dry natron, a naturally occurring salt mixture with a large proportion of sodium carbonate, followed by wrapping in multiple layers of resin-infiltrated bandages (David and Tapp 1984). In some instances, for example Punn II (170 B.C. \pm 70 years), resin was also poured into the abdominal, thoracic and cranial cavities (Cockburn *et al.* 1975). It is significant that Punn II was especially well-preserved; immediately on unwrapping, swabs taken from the body cavity proved to be sterile and intact proteins of a molecular weight corresponding to gamma globulin were recovered (Cockburn *et al.* 1975). In contrast, little resin was found in the body of Manchester mummy 1770 (Greek/Roman period) which was described as 'in an advanced state of decomposition' (Tapp 1979). The suggestion, therefore, is that the extensive use of resin is at least associated with reduced decay. The importance of dehydration in preservation is further illustrated by the experimental mummification of rats: after reaching a threshold level of dehydration (for rats this is after 30–40 days in dry natron), further decay was minimal even when the carcass was returned to 'normal' conditions (Garner 1979).

Surveying the available evidence suggests that both dehydration and the antibiotic properties of resin may contribute to the exceptional preservation of amber fossils; assessing their relative roles in decay limitation, however, awaits further investigation.

Further work will explore the application of the soft-part preservation and histological detail described here. There is no reason to assume that the preservation of amber inclusions does not extend to the many insects fossilized in amber of diverse geological ages and geographical origin; these faunas may further increase our knowledge of soft-part evolution. The rich biota trapped in amber of various geological ages represents a considerable resource, as yet largely untapped. Further work, including the development of improved sectioning techniques, will, it is hoped, enable detailed study of comparative histology in the evolution of insects.

Acknowledgements. I thank Simon Conway Morris, Sue Rigby, Lesley Dowling, Ken Harvey, Roger Northfield, David Newling, Ben Harris and Jake Brodzinsky. Two anonymous reviewers provided helpful comments. This work was funded by a NERC postgraduate grant, a Gateway Corporation scholarship and by the Cowper Reed fund of the Department of Earth Sciences, Cambridge University. Terry Doyle (Keele University) drafted Text-figure 1.

REFERENCES

- ALEKSEYEV, V. N. and RASNITSYN, A. P. 1981. [Late Cretaceous Magaspilidae (Hymenoptera) from amber of Taimyr]. *Palaeontological Journal*, **15**, 124–128. [Translated from *Paleontologicheskii Zhurnal*, **4**, 127–130].
- ALEXANDER, R. MCNEILL 1979. *The invertebrates*. Cambridge University Press, Cambridge, 562 pp.
- ALLISON, P. A. 1988. The role of anoxia in the decay and mineralization of proteinaceous macrofossils. *Paleobiology*, **14**, 139–154.
- ARRHENIUS, S. and LANGENHEIM, J. H. 1983. Inhibitory effects of *Hymenaea* and *Copaifera* leaf resins on the leaf fungus *Pestalotia*. *Biochemical and Systematic Ecology*, **11**, 361–366.
- BALFOUR-BROWNE, F. 1944. The proventriculus of the Coleoptera (Adephaga) and other insects – a study in evolution. *Journal of the Royal Microscopical Society*, **64**, 68–118.
- BARONI-URBANI, C. and SAUNDERS, J. B. 1982. The fauna of the Dominican Republic amber: the present status of knowledge. 213–223. In *Proceedings of the 9th Caribbean Conference, Santo Domingo 1980*, 711 pp.
- BERNER, R. A. and CANFIELD, D. E. 1989. A new model for atmospheric oxygen over Phanerozoic time. *American Journal of Science*, **289**, 333–361.
- and LANDIS, G. P. 1988. Gas bubbles in fossil amber as possible indicators of the major gas composition of ancient air. *Science*, **239**, 1406–1409.
- BROUWER, S. B. and BROUWER, P. A. 1980. Geología de la región ambarífera oriental de la República Dominicana. 303–333. In *Proceedings of the 9th Caribbean Conference, Santo Domingo 1980*, 711 pp.
- BRUES, C. T. 1951. Insects in amber. *Scientific American*, **185**, 56–61.
- BUTTERFIELD, N. J. 1990. Organic preservation of non-mineralizing organisms and the taphonomy of the Burgess shale. *Paleobiology*, **16**, 272–286.
- CHAMPETIER, Y., MADRE, M., SAMAMA, J. C. and TAVERES, I. 1982. Localisation de l'ambre au sein des séquences à lignites en République Dominicaine. 277–280. In *Proceedings of the 9th Caribbean Conference, Santo Domingo 1980*, 711 pp.
- CHAPMAN, R. F. 1982. *The insects, structure and function*. Hodder and Stoughton, London, 919 pp.
- COCKBURN, A., BARRACO, R. A., REYMAN, T. A. and PECK, W. H. 1975. Autopsy of an Egyptian mummy. *Science*, **187**, 1155–1160.
- CROWSON, R. A. 1981. *The biology of the Coleoptera*. Academic Press, London and New York, 802 pp.
- DAVID, R. and TAPP, E. 1984. *Evidence embalmed. Modern medicine and the mummies of ancient Egypt*. Manchester University Press, Manchester, 175 pp.
- EBERLE, W., HIRDES, W., MUFF, R. and PELAEZ, M. 1982. The geology of the Cordillera Septentrional (Dominican Republic). 619–629. In *Proceedings of the 9th Caribbean Conference, Santo Domingo 1980*, 711 pp.
- FRIS, E. M., CHALONER, W. G. and CRANE, P. R. 1987. *The origins of angiosperms and their biological consequences*. Cambridge University Press, Cambridge, 358 pp.
- GARNER, R. 1979. Experimental mummification. 19–24. In DAVID, A. R. (ed.). *The Manchester Museum Mummy Project: multidisciplinary research on Ancient Egyptian mummified remains*. Manchester University Press, Manchester, 160 pp.
- HENWOOD, A. A. 1992. Exceptional preservation of dipteran flight muscle and the taphonomy of insects in amber. *Palaio*, **7**, 203–212.
- HOPFENBERG, H. B., WHITCHEY, L. C. and POINAR, G. O., jr 1988. Is the air in amber ancient? *Science*, **242**, 717–718.
- HORIBE, Y. and CRAIG, H. 1988. Is the air in amber ancient? *Science*, **242**, 720–721.
- HUEBER, F. M. and LANGENHEIM, J. H. 1986. Dominican amber tree had African ancestors. *Geotimes*, **31**, 8.
- KEIL, T. A. and STEINBRECHT, R. A. 1982. Mechanosensitive and olfactory sensilla of insects. 477–516. In KING, R. C. and AKAI, H. (eds). *Insect ultrastructure*. Plenum Press, New York and London, 770 pp.
- KEILBACH, R. 1982. Bibliographie und Liste der Arten Tierischer Einschlüsse in fossilen Harzen sowie ihrer Aufbewahrungsorte. *Deutsche Entomologische Zeitschrift, Neue Folge*, **29**, Heft 1–3, 129–286.
- KIDWELL, S. M. and BAUMILLER, T. 1990. Experimental disintegration of regular echinoids: role of temperature, oxygen and decay thresholds. *Paleobiology*, **16**, 247–271.
- KLUMP, J. B. and GARRELS, R. M. 1986. Modelling atmospheric oxygen in the global sedimentary redox cycle. *American Journal of Science*, **286**, 337–360.
- LAMBERT, J. B., FRYE, J. S. and POINAR, G. O., jr 1985. Amber from the Dominican Republic: analysis by nuclear magnetic resonance spectroscopy. *Archeometry*, **27**, 43–51.
- LANGENHEIM, J. H. 1964. Present status of botanical studies of ambers. *Botanical Museum Leaflets, Harvard University*, **20**, 225–287.

- LANGENHEIM, J. H. and BECK, C. W. 1965. Infrared spectra as a means of determining botanical sources of amber. *Science*, **44**, 52–55.
- LARSSON, S. G. 1978. *Baltic amber – a palaeobiological study. Entomonograph volume 1*. Scandinavian Science Press Ltd, Klampenborg, Denmark, 192 pp.
- LEWIS, J. F. and DRAPER, G. 1990. Geology and tectonic evolution of the northern Caribbean margin. 77–140. In DENG, G. and CASE, J. E. (eds). *The Geology of North America, volume H, the Caribbean region*. The Geological Society of America, Boulder, Colorado, 528 pp.
- MARTILL, D. M. 1990. Macromolecular resolution of fossilized muscle tissue from an elaphomorph fish. *Nature*, **346**, 171–172.
- PENDERSON, T. F. and CALVERT, S. E. 1990. Anoxia vs productivity: what controls the formation of organic-carbon-rich sediments and sedimentary rocks? *Bulletin of the American Association of Petroleum Geologists*, **74**, 458–466.
- POINAR, G. O. jr 1988. Hair in Dominican amber: evidence for Tertiary land mammals in the Antilles. *Experientia*, **44**, 88–89.
- and CANNATELLA, D. 1987. Upper Eocene frog from the Dominican Republic and its implication for Caribbean biogeography. *Science*, **237**, 1215.
- and HESS, R. 1985. Preservative qualities of amber. *Baltic Studies*, **16**, 222–230.
- — 1982. Ultrastructure of 40-million-year-old insect tissue. *Science*, **215**, 1241–1242.
- and SINGER, R. 1990. Upper Eocene gilled mushroom from the Dominican Republic. *Science*, **248**, 1099–1101.
- PRINGLE, J. W. S. 1980. The evolution of fibrillar muscle in insects. *Journal of Experimental Biology*, **94**, 1–14.
- REDMOND, B. 1980. The Tertiary of the central Cordillera Septentrional. 630–638. In *Proceedings of the 9th Caribbean Conference, Santo Domingo 1980*, 711 pp.
- RICHARDS, A. G. 1951. *The integument of arthropods*. University of Minnesota Press, Minneapolis, 163 pp.
- SANDERSON, M. W. and FARR, T. H. 1960. Amber with insect and plant inclusions from the Dominican Republic. *Science*, **131**, 1313.
- SAUNDERS, J. B., JUNG, P. and BIJU-DUVAL, B. 1982. Neogene paleontology in the northern Dominican Republic. 1. Field surveys, lithology, environment and age. *Bulletins of American Paleontology*, **89**, 1–79.
- SCHLEE, D. 1980. *Bernstein Raritäten: Farben, Strukturen, Fossilien, Handwerk. Aus der Bernsteinsammlung des Staatlichen Museums für Naturkunde in Stuttgart*. Staatlichen Museum für Naturkunde, Stuttgart, 88 pp.
- 1990. Bernstein Kabinett. *Stuttgarter Beiträge zur Naturkunde Series C*, **28**, 1–128.
- and DIETRICH, H.-G. 1970. Insektenführender Bernstein aus der Unterkreide des Libanon. *Neues Jahrbuch für Geologie und Paläontologie, Monatshefte*, 1970, 40–50.
- SHACKLETON, N. J. 1985. Oceanic carbon isotope constraints on oxygen and carbon dioxide in the Cenozoic atmosphere. *Memoirs of the American Geophysical Union*, **32**, 412–417.
- SMITH, D. S. 1961. The structure of insect fibrillar flight muscle. A study made with special reference to the membrane system of the fibre. *Journal of Biophysical and Biochemical Cytology*, **10**, supplement, 123–58.
- 1966. The organization and function of the sarcoplasmic reticulum and T-system of muscle cells. *Progress in Biophysical and Molecular Biology*, **16**, 107–142.
- TAPP, E. 1979. The unwrapping of a mummy. 83–94. In DAVID, A. R. (ed.). *The Manchester Museum mummy project: multidisciplinary research on ancient Egyptian mummified remains*. Manchester University Press, Manchester, 160 pp.
- VOIGT, E. 1988. Preservation of soft tissues in the Eocene lignite of the Geiseltal near Halles. *Courier Forschungsinstitut – Institut Senckenberg*, **107**, 325–343.
- WEIS-FOGH, T. 1964. Diffusion in insect wing muscle, the most active tissue known. *Journal of Experimental Biology*, **41**, 229–256.
- ZOETEN, R. DE 1988. Structure and stratigraphy of the Central Cordillera Septentrional, Dominican Republic. Unpublished M.A. thesis, University of Texas at Austin.

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Typescript received 25 April 1991

Revised typescript received 4 April 1992